

Applicant : Michael A. Apicella et al.
Serial No. : 09/574,460
Filed : May 18, 2000
Page : 2 of 7

Attorney's Docket No.: 17023-004001/98010

Amendments to the Claims:

1-29. (Cancelled)

30. (Currently amended) A process for the production of a *Haemophilus influenzae*-specific lipooligosaccharide (LOS) which comprises the steps of:

(a) growing in a culture medium gram-negative bacteria comprising (i) a core lipid structure containing a terminal heptose and (ii) a DNA sequence comprising a an Undecapaprenyl-phosphate (UDP) N-acetyl glucosamine (GlcNAc):Undecaprenol GlcNAc-1 phosphate transferase rfe (rfe) gene, and (iii) an isolated DNA sequence encoding comprising a lipooligosaccharide-synthesis gene G (lsgG) gene product (LsgG) from *Haemophilus influenzae*, ~~wherein lsgG encodes LsgG, and~~ wherein the *rfe* is regulated by LsgG such that a *H. influenzae*-specific LOS is synthesized by the addition of an acceptor molecule to the terminal heptose molecule; and

(b) recovering the *H. influenzae*-specific LOS from the culture medium.

31. (Previously presented) The process of claim 30, wherein the bacteria are *Escherichia coli*.

32. (Previously presented) The process of claim 31, wherein the bacteria are *Escherichia coli* K-12 strain JM 109.

33. (Previously presented) The process of claim 30, wherein the bacteria are *Salmonella minnesota*.

34. (Previously presented) The process of claim 30, wherein the acceptor molecule is N-acetylglucosamine.

35. (Previously presented) The process of claim 30, wherein the *rfe* gene is from *Haemophilus influenzae*.

Applicant : Michael A. Apicella et al.
Serial No. : 09/574,460
Filed : May 18, 2000
Page : 3 of 7

Attorney's Docket No.: 17023-004001/98010

36. (Previously presented) The process of claim 30, wherein the DNA sequence comprising a *rfe* gene is part of the gram-negative bacterial genome.

37. (Currently amended) The process of claim 30, wherein the isolated DNA sequence encoding ~~comprising~~ the *lsgG* is contained in a vector.

38. (Previously presented) The process of claim 30, wherein the bacteria further comprise a glycosyltransferase.

39. (Currently amended) A process for the production of a complex carbohydrate comprising the steps of:

- (a) growing in a culture medium gram-negative bacteria comprising (i) a core lipid structure containing a terminal heptose and (ii) a DNA sequence comprising a an Undecapaprenyl-phosphate (UDP) N-acetyl glucosamine (GlcNAc):Undecaprenol GlcNAc-1 phosphate transferase ~~rfe~~ (*rfe*) gene, and (iii) an isolated DNA sequence ~~comprising~~ encoding a liposaccharide-synthesis gene G (*lsgG*) gene product (LsgG) from *Haemophilus influenzae*, ~~wherein *lsgG* encodes LsgG, and wherein the *rfe* gene is regulated by LsgG such that a complex carbohydrate is synthesized by the addition of an acceptor molecule to the heptose molecule; and~~
 - (b) recovering the complex carbohydrate from the culture medium.

40. (Previously presented) The process of claim 39, wherein the bacteria are *Escherichia coli*.

41. (Previously presented) The process of claim 40, wherein the bacteria are *Escherichia coli* K-12 strain JM 109.

42. (Previously presented) The process of claim 39, wherein the bacteria are *Salmonella minnesota*.

43. (Previously presented) The process of claim 39, wherein the acceptor molecule is N-acetylglucosamine.

Applicant : Michael A. Apicella et al.
Serial No. : 09/574,460
Filed : May 18, 2000
Page : 4 of 7

Attorney's Docket No.: 17023-004001/98010

44. (Previously presented) The process of claim 39, wherein the *rfe* gene is from *Haemophilus influenzae*.
45. (Previously presented) The process of claim 39, wherein the DNA sequence comprising a *rfe* gene is part of the gram-negative bacterial genome.
46. (Previously presented) The process of claim 39, wherein the isolated DNA sequence comprising the *lsgG* is contained in a vector.
47. (Previously presented) The process of claim 39, wherein the bacteria further comprise a glycosyltransferase.
48. (Currently amended) A method of modifying a terminal heptose of a lipopolysaccharide (LPS) or lipooligosaccharide (LOS) core structure of a gram-negative bacterial species containing a an Undecapaprenyl-phosphate (UDP) N-acetyl glucosamine (GlcNAc):Undecaprenol GlcNAc-1 phosphate transferase *rfe* (*rfe*) gene comprising regulating the *rfe* gene with a protein encoded by an isolated lipooligosaccharide-synthesis gene *G lsgG* (*lsgG*) gene from *Haemophilus influenzae* such that an N-acetyl glucosamine is added onto the terminal heptose.
49. (Previously presented) The method of claim 48 wherein the bacteria are *Escherichia coli*.
50. (Previously presented) The method of claim 49, wherein the bacteria are *Escherichia coli* K-12 strain JM 109.
51. (Previously presented) The method of claim 48, wherein the bacteria are *Salmonella minnesota*.

Applicant : Michael A. Apicella et al.
Serial No. : 09/574,460
Filed : May 18, 2000
Page : 5 of 7

Attorney's Docket No.: 17023-004001 / 98010

52. (Previously presented) The method of claim 48, wherein the *rfe* gene is from *Haemophilus influenzae*.
53. (Previously presented) The method of claim 48, wherein the *rfe* gene is part of the gram-negative bacterial genome.
54. (Previously presented) The method of claim 48, wherein the isolated *lsgG* gene is contained in a vector.
55. (Previously presented) The method of claim 48, wherein the bacteria further comprise a glycosyltransferase.